भारतीय मानक
Indian Standard

IS 3025 (Part 68) : 2019

जल एवं अपशिष्ट जल के नमूने लेने तथा परीक्षण (भौतिक एवं रसायन) की पद्धतियां

भाग 68 अनायनिक सर्फैक्टैन्ट्स

Methods of Sampling and Test (Physical and Chemical) for Water and Wastewater

Part 68 Anionic Surfactants

ICS 13.060.50

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FOREWORD

This Indian Standard (Part 68) was adopted by the Bureau of Indian Standards, after the draft finalized by the Water Quality Sectional Committee had been approved by the Chemical Division Council.

Surfactants enter water and wastewater mainly by discharge of aqueous wastes from households and industrial laundering and/or other cleansing operations. A surfactant is a single molecule which contains a hydrophic group and a hydrophilic group. Due to this bivalent nature, these molecules tend to congregate at interfaces between aqueous phase and other phases such as air/oil/dust; and due to this congregation foaming, emulsification and particle suspension occurs.

Hydrophobic group of the surfactants are generally hydrocarbon radical (R) containing about 10-20 carbon atoms. The hydrophilic groups can be of two types; those that ionize in water and those that do not ionize in water. The ionizing groups can further be of two types:

a) Anionic (carrying negative charge) — for example, $(RSO_3^-)Na^+$; or

b) Cationic (carrying positive charge) — For example, $(RMe_3N)^+Cl^-$.

Anionic surfactants are used in laundry and hand dishwashing detergents, household cleaners, and personal cleansing products. Linear alkylbenzenesulfonate, alcohol ethoxysulfates, alkyl sulfates, and soap are common anionic surfactants. This standard prescribes test method for determination of anionic surfactant.

The method is applicable to drinking water, surface water as well as waste water, for example for the determination of the primary degradation of surfactants under investigation in test systems containing natural or synthetic waste water. It applies for both laboratory scale and technical waste water treatment plants. This standard is technically equivalent to ISO 7875-1 : 1996.

The composition of the Committee responsible for the formulation of this standard is given at Annex A.

In reporting the results of a test or analysis in accordance with this standard, if the final value observed or calculated, is to be rounded off, it shall be done in accordance with IS 2 : 1960 “Rules for rounding off numerical values (*revised*)”.

Indian Standard

METHODS OF SAMPLING AND TEST (PHYSICAL AND CHEMICAL) FOR WATER AND WASTEWATER

PART 68 ANIONIC SURFACTANTS

1 SCOPE

1.1 This standard (Part 68) prescribes a spectrometric method of test for the determination of anioninc surfactants by measurement of the methylene blue index (MBAS) in aqueous media.

1.2 This method is applicable to a range of concentrations from 0.1 mg/l to 5.0 mg/l and the limit of detection is about 0.05 mg/l for solutions of standard surfactants in distilled water.

2 REFERENCES

The standards listed below contain provisions which through reference in this text, constitute provisions of this standard. At the time of publications, the editions indicated were valid. All standards are subject to revision, and parties to agreements based on this standard are encouraged to investigate the possibility of applying the most recent editions of the standards indicated below:

IS No.	*Title*
1070 : 1992	Reagent grade water
3025 (Part 1) : 1987	Methods of sampling and test (physical and chemical) for water and wastewater: Part 1 Sampling

3 PRINCIPLE

3.1 Methylene Blue forms a complex with anionic surfactants in an alkaline medium. Interference from the matrix is eliminated by extracting the anionic surfactant-methylene blue complex with chloroform and then treated with acidic methylene blue solution. Absorbance of the separated organic phase is measured at the maximum absorption wavelength of 650 nm and evaluated by means of calibration curve.

3.2 For reasons of purity and stability, the preferred standard is dodecyl benzene sulfonic acid methyl ester (tetrapropylene type, of relative molecular mass 340), although other calibration standards may be used (*see* the note to **4.11**). The calibration standard is prepared from the standard dodecyl benzene sulfonic acid ester after saponification to the sodium salt. Calculation of the MBAS index as sodium dodecyl benzene sulfonate is given in **8.1**.

3.3 Under the experimental conditions, sulfonates and sulfates are the compounds chiefly measured, but some positive and negative interferences may occur (*see* **9**). In the case of effluents originating from municipal waste water treatment plants, the MBAS index comprises not only synthetic but also, to a considerable extent, natural anionic surface active substances.

4 REAGENTS

During the analysis, unless otherwise stated, use only reagents of recognized analytical grade and only distilled water or water of equivalent purity (*see* IS 1070).

4.1 Sodium Chloride (NaCl)

4.2 Ethyl Acetate ($C_4H_8O_2$)

Freshly distilled.

CAUTION — Ethyl acetate is flammable and toxic upon inhalation/ingestion. Irritant for skin/eye.

4.3 Chloroform ($CHCl_3$)

CAUTION — Chloroform is a suspected carcinogen.

If necessary [for example, if it gives rise to high results in blank test (**7.2**)] purify the chloroform by filtration through aluminum oxide (Al_2O_3) (neutral grade, W 200).

NOTE — Due to the toxicity of chloroform, it would be preferable to replace it by another solvent.

4.4 Ethanol (C_2H_5OH)

95 percent (*v*/*v*)

4.5 Methanol (CH_3OH)

Freshly distilled. In order to avoid high results in blank tests (**7.2**) store in glass bottle.

CAUTION — Methanol is highly flammable and toxic

4.6 Sulfuric Acid (H_2SO_4)

0.5 mol/l solution.

4.7 Ethanolic Sodium Hydroxide (NaOH),

0.1 mol/l solution in ethanol. Dissolve 4 g of sodium hydroxide pellets in ethanol (**4.4**) and dilute to 1 000 ml with the same ethanol.

4.8 Methylene Blue (Neutral Solution)

NOTE — The solid methylene blue used should be the purest available.

Dissolve 0.350 g of methylene blue in water and dilute to 1 000 ml.

Prepare the solution at least 24 h before use and this solution is stable for at least 2 weeks.

The absorbance of the chloroform phase of the blank test (*see* **7.2**), measured against chloroform, shall not exceed 0.2 per 10 mm of optical path length at 650 nm. In the case of higher absorbance during the blank test, use other batches of methylene blue and/or purify the methylene blue solution by extraction as follows:

Place the methylene blue solution (**4.8**) in a suitably large separating funnel. For each 100 ml of methylene blue solution, add 200 ml of the buffer solution (**4.10**) and 200 ml of chloroform (**4.3**). Shake for 30 s and allow to separate. Run off the chloroform layer as completely as possible and rinse the aqueous layer without shaking with 60 ml of chloroform for each 100 ml of methylene blue solution. Repeat the extraction and rinse as before. Discard the chloroform extracts, collect for reuse after treatment.

4.9 Methylene Blue (Acidic Solution)

Dissolve 0.350 g of methylene blue in 500 ml of water and add 6.50 ml of sulfuric acid (ρ = 1.84 g/ml). Dilute with water to 1 000 ml after mixing.

Prepare the solution at least 24 h before use.

The absorbance of the chloroform phase of the blank test (*see* **7.2**), measured against chloroform, shall not exceed 0.02 per 10 mm of optical path length at 650 nm. In the case of higher blank absorbances, either wash the methylene blue solution twice with chloroform for purification (*see* **4.8**) or use other batches of methylene blue.

4.10 Buffer Solution (Of pH 10)

4.10.1 Dissolve 24 g of sodium hydrogen carbonate ($NaHCO_3$) and 27 g of anhydrous sodium carbonate (Na_2CO_3) in water and dilute to 1 000 ml.

4.10.2 Alternatively, especially for very hard water, the buffer solution prepared in **4.10.2.3** may be used.

4.10.2.1 *Disodium tetraborate ($Na_2B_4O_7.10H_2O$), 0.05 mol/l solution*

Dissolve 19 g of disodium tetraborate decahydrate in 1 000 ml of water.

This solution is stable for at least 2 weeks if stored in a stoppered glass bottle.

4.10.2.2 *Sodium hydroxide (NaOH), 0.1 mol/l solution*

Dissolve 4 g of sodium hydroxide pellets in 1 000 ml of water.

This solution is stable for at least 2 weeks if stored in a glass bottle with a polyethylene stopper.

4.10.2.3 *Borate, alkaline solution*

Mix equal volumes of disodium tetraborate solution (**4.10.2.1**) and sodium hydroxide solution (**4.10.2.2**).

This solution is stable for at least 1 week if stored in a glass bottle with a polyethylene stopper.

4.11 Dodecylbenzene Sulfonic Acid Methyl Ester (Tetrapropylene Type) ($C_{19}H_{32}O_3S$), Stock Standard Solution

Weigh, preferably from a weighing pipette, 400 mg of dodecylbenzene sulfonic acid methyl ester to the nearest 0.1 mg, into a round-bottomed flask, and add 50 ml of ethanolic sodium hydroxide solution (**4.7**) and some anti-bumping granules. Attach the reflux condenser and boil for 1 h. After cooling, rinse the condenser and the ground-glass joint with about 30 ml of ethanol (**4.4**) and add the washings to the contents of the flask. Neutralize the solution with sulfuric acid (**4.6**) against phenolphthalein (**4.12**) until it becomes colourless. Transfer the solution to a 1 000 ml one-mark volumetric flask, dilute to the mark with water and mix.

This standard solution is stable for at least 6 months.

NOTE — Although the dodecylbenzene sulfonic acid methyl ester is preferable as it is a guaranteed nonhygroscopic standard, the calibration graph (*see* **7.3**) may alternatively be established with the aid of commercially available sodium salt of dodecane-1 sulfonic acid ($C_{12}H_{25}NaO_3S$), dodecane-1 sulfuric acid ($C_{12}H_{25}NaO_4S$) or dioctyl sulfosuccinic acid ($C_{20}H_{37}NaO_7S$).

4.12 Phenolphthalein (Indicator Solution)

Dissolve 1.0 g of phenolphthalein in 50 ml of ethanol (**4.4**) and add, while stirring continuously, 50 ml of water. Filter off any precipitate that forms.

5 APPARATUS

Ordinary laboratory equipment and the following:

5.1 pH-meter

With suitable electrodes made from glass.

5.2 UV-VIS Spectrometer,

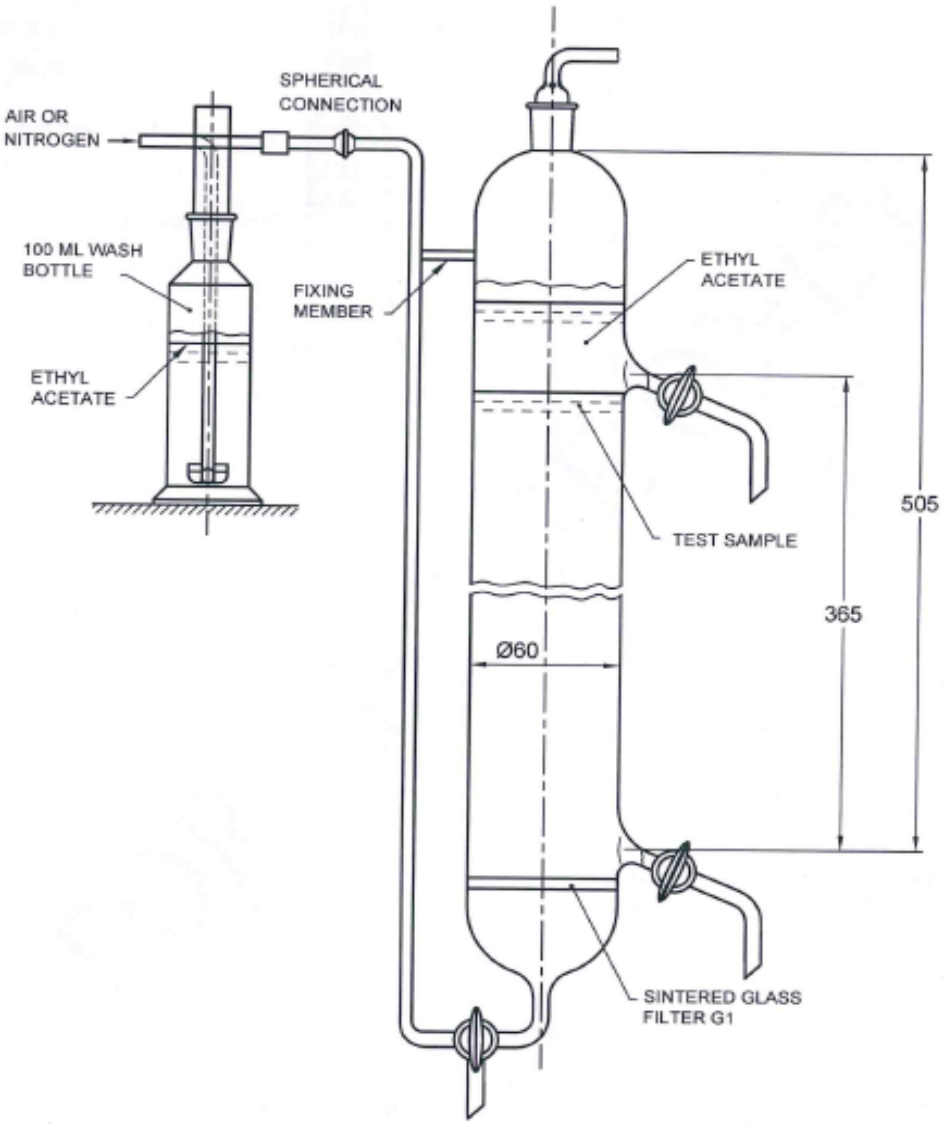
Capable of measurement at 650 nm, equipped with quartz cuvettes of optional path lengths 10 mm and 50 mm.

5.3 Gas-stripping Apparatus (*see* Fig. 1**)**

Which is commercially available of capacity 1 litre.

NOTES

1 To make cleaning easier, the apparatus should preferably be equipped with a spherical connection under the stripping funnel. The fixing member should also be divisible.



All dimensions are in millimeters.

FIG. 1 GAS STRIPPING APPARATUS

2 During preliminary cleaning, all glassware should be washed thoroughly with water and then with ethanolic hydrochloric acid about 10 percent (*v/v*) and subsequently rinsed with water.

6 SAMPLING AND SAMPLES

Instructions prescribed for sampling in IS 3025 (Part 1) is recommended to be followed.

Do not withdraw samples through a foam layer. Use clean glass bottles, previously washed with methanol (**4.5**) for sampling and storage. Cooling to 4°C is recommended for preservation over short periods. Consider the addition of a preservative if the sample is to be kept for more than 21 h. The addition of 1percent (v/*v*) of a 40 percent (*v/v*) formaldehyde solution is suitable for periods up to 4 days while saturating with chloroform is suitable for periods up to 8 days.

NOTE — Test samples should normally be free of suspended matter which can be separated by centrifugation, however it should be appreciated that, as a result of such separation, surfactant adsorbed on suspended matter will not be determined.

7 PROCEDURE

7.1 Concentration and Separation of the Surfactant

For all types of water with known matrices and/or free of interferences proceed according to **7.4**. For determination of the total amount of MBAS in the presence of solids, also proceed according to **7.4**, although quantitative recovery is not guaranteed due to sorption effects. For analysis of the amount of dissolved MBAS, use the following concentration and separation procedure.

Non-surfactant methylene blue active substances can cause errors in the determination of the methylene blue index. In surface water and other types of water with unknown composition or known to contain interfering compounds, separate the surfactants by stripping (solvent sublation). Stripping is also recommended for concentrating small amounts of surfactants from water samples. Separate suspended matter by centrifugation, but note that adsorbed surfactant on suspended matter will not be determined.

Place a measured quantity of the test sample, up to 1 000 ml, in the gas-stripping apparatus (**5.3**).

Install the sripping apparatus in well-ventilated hood to carry off ethyl acetate vapour.

Separation is improved by the addition of sodium chloride. If the test sample volume exceeds 500 ml, add 100 g of solid sodium chloride and dissolve by passing nitrogen gas or air through it. If a smaller test sample volume is used, dissolve 100 g of sodium chloride in 400 ml of distilled water and add the solution, to the test sample.

If necessary, add distilled water to bring the sample surface up to the level of the upper stopcock (*see* Fig. 1). Add 100 ml of ethyl acetate (**4.2**). Fill two-thirds of the volume of the wash bottle in the gas line (*see* Fig. 1) with ethyl acetate. Pass a gas stream of 20 l/h to 50 l/h through the gas-stripping apparatus. Use of a flowmeter with variable area is recommended. Adjust the gas flow in such a way that the phases remain separate and no turbulence is produced at the interface. Significant mixing of the phases and consequent solution of ethyl acetate in the water is to be avoided. Stop the gas flow after 5 min.

If a loss of more than 20 percent (*v/v*) of the organic phase has occurred due to evaporation/dissolution in the water phase, discard the test sample and carry out separation using new test sample.

Run off the organic phase completely into a separating funnel. Return any water in the separating funnel (there should only be a few mililitres) to the gas-stripping apparatus.

Filter the ethyl acetate solution through a dry qualitative gas-filter paper into a 250 ml flask. Add a further 100 ml of ethyl acetate to the gas-stripping apparatus and again pass nitrogen or air through it for 5 min. Separate the organic layer described above, using the same separating funnel, filter, and add it to the first portion. Rinse the filter paper and funnel with 25 ml of ethyl acetate. Evaporate the ethyl acetate solution on a water-bath under a hood. To speed up the process, direct a gentle air stream over the surface of the solution.

Dissolve the dry residue in about 5 ml of methanol (**4.5**) and 50 ml of water. Transfer the solution quantitatively to a 100 ml volumetric flask and make up the volume with water.

7.2 Blank Test

With each series of test samples, carry out a blank test in parallel with the determination, using the zero member of the set of calibration solutions (*see* **7.3**).

Subtract the interpolated absorbance, A_0, from the absorbance, A_1 of the test sample. Under the given conditions the absorbance, A_0 of the blank test shall not exceed 0.02 for 10 mm optical path length, otherwise the equipment and the reagent shall be checked carefully for any contamination.

7.3 Calibration

From the stock standard solution (**4.11**), prepare a working standard solution by transferring 25 ml with a pipette to a 500 ml volumetric flask, dilute to the mark with water and mix.

The mass concentration of MBAS, ρ_x, in mg/ml, of this working standard solution is given by the equation:

$$p_x \frac{mf_1}{V}$$

where,

m = mass of the MBAS (as ester) used for preparation of the stock standard solution according to 4.11 (mg),

f_1 = conversion factor from ester to MBAS, sodium salt of dodecylbenzene sulfonate, here f_1 = 1.023 5 (*see* Table 1), or

V = volume correction factor (in ml), here V = 20 000 ml.

Place 0.0 ml (the zero member); 1.0 ml, 2.0 ml, 4.0 ml, 6.0 ml and 8.0 ml of the working standard in a series of separating funnels of capacity 250 ml, dilute with water to 100 ml and continue as described in **7.4**.

Measure the absorbance of each of the set of the calibration solutions, including the zero member, at a wavelength of 650 nm in cells of optical path lengths of 10 mm or 50 mm. Prepare a calibration graph by plotting the absorbance against the mass, in micrograms, of surfactant contained in the calibration solution and subtract the interpolated absorbances (intersection with the ordinate) of the blank from the absorbances, A_1, of each calibration solution (**8.1**).

Calibrate once or twice a month or whenever new batches of chemicals are used.

If the calibration is carried out with one of the alternative surfactants (*see* **4.11**), use the conversion factors f_1 shown in Table 1.

Table 1 Conversion Factors of Surfactants

Sl No.	Surfactant	Conversion Factor, f_1
(1)	(2)	(3)
(i)	Dodecylbenzene sulfonic acid, sodium salt	1.000
(ii)	Dodecane-1-sulfonic acid, sodium salt	0.781 6
(iii)	Dodecan-1-sulfuric acid, sodium salt	0.827 6
(iv)	Dioctyl sulfosuccinic acid, sodium salt	1.276 0

7.4 Determination

Transfer a measured volume of the test sample, if necessary treated according to **7.1**, into a separating funnel such that MBAS content in the test sample should be between 20 µg and 200 µg. In the lower MBAS range, a test portion of up to 100 ml may be used; if the volume of the test portion is less than 100 ml, dilute with water to 100 ml. Add 5.0 ml of neutral methylene blue solution (**4.8**), 10 ml of buffer solution (**4.10**) (not necessary if a pre-extracted methylene blue solution is used), and 15 ml of chloroform (**4.3**). Shake evenly and gently for 1 min.. Allow the layers to separate as completely as possible and swirl the funnel to dislodge droplets from the sides of the funnel.

Allow to settle for 2 min., then run (as much as possible) of the chloroform layer into a second separating funnel, containing 110 ml of water and 5.0 ml of acidic methylene blue solution (**4.9**). Shake uniformly, but not too vigorously for 1 minutes as previously described. Filter the chloroform layer through a cotton or glass wool filter wetted with chloroform (**4.3**) into a 50 ml volumetric flask.

NOTE — On cotton wool, some absorption of surfactants may take place; on glass wool, water may not be absorbed completely.

Repeat the extraction using 10 ml portion of chloroform (**4.3**) added to the first separating funnel of the alkaline and then acid solutions for the extraction as explained earlier. Separate the chloroform and filter it, through the same filter, into the volumetric flask. Repeat the extraction using a further 10 ml portion of chloroform and filter that into the 50 ml volumetric flask. Dilute to the mark with chloroform and mix.

For each batch of test samples, carry out the complete extraction for a blank determination on 100 ml of water and on one of the calibration solutions (*see* **7.3**).

Before each determination, shake the contents of the volumetric flask, rinse the optical cell of the spectrometer (**5.2**) three times with the solution in respective flask, and then fill the cell.

Measure the absorbances for test samples, calibration solutions and the blank test with a spectrometer at a wavelength of 650 nm in cells of optical path lengths of 10 mm or 50 mm against chloroform. Comparison measurements on standards shall be made in the same size of cells. Wash out the cells with chloroform after each reading.

Check the cell error frequently by measuring the absorbance difference when chloroform is used in both cells and correct for any error. If this error increases, clean the cell by immersion in nitric acid, rinsing with water, and drying with acetone and chloroform. Mark one cell and reserve for the reference chloroform.

If the absorbance of the test solution of the sample when measured in cells of optical path length 10 mm is less than 0.1, repeat readings of calibration solutions, blank test and test sample in 40 mm or 50 mm cells.

If the standard solutions run with the sample batch differ significantly from the calibration graph value, repeat the procedure with all samples and a full set of calibration solutions.

8 EXPRESSION OF RESULTS

8.1 Calculation

Calculate the MBAS index as mass concentration, ρv, expressed in micrograms per milliliter, calculated as the sodium salt of dodecyl benzene sulfonic acid, using the equation:

$$\rho_v = \frac{f_2 \times (A_1 - A_0)}{V_0}$$

where,

A_1 = absorbance of test sample,

A_0 = absorbance of blank,

f_2 = calibration factor representing the mass, in micrograms of MBAS (calculated as the sodium salt of dodecyl benzene sulfonic acid) under which the given conditions yields on absorbance of 1.000 (evaluated from calibration graph), and

V_0 = volume, in millilitres, of the test portion taken for analysis according to **7.4**. If the sample was diluted, this shall be accounted for (*see* **7.1**). If the sample was sublated, V_0 represents the 100 ml obtained according to **7.1**.

Alternatively, determine the MBAS mass concentration from the calibration graph (*see* **7.3**). The mass concentration is calculated from the mass of MBAS in the test portion, taken from the calibration graph, and its volume.

8.2 Precision

The precision, P, of the method can be expressed as

$$P = 0.107\rho_Y + 0.008$$

Where, ρ_Y is the mass concentration, expressed in micrograms per milliliter, of MBAS.

At 0.1 µg/ml, the relative standard deviation, *s*, was calculated to be *s* = ± 19 percent.

9 INTERFERENCES

MBAS values which are falsely low can be obtained in the presence of cationic substances such as quaternary ammonium compounds and proteins which form compounds with anionic surfactants. For example, if the sample contains anionic as well as cationic surfactants, these may form stable complexes which will not react with methylene blue.

MBAS values which are falsely high may be caused by substances other than anionic surfactants forming compounds with methylene blue which are soluble in chloroform. This interference, is minimized by stripping the surfactants from the sample into ethyl acetate, thus separating them from non-surface active substances (*see* **7.1**).

In theory, any compound containing a single strong anionic grouping and a hydrophobic part is capable of forming an extractable ion-association compound with the methylene blue cation. Organic sulfates, sulfonates, carboxylates, phenols, and inorganic anions such as isocyanate, nitrate, thiocyanate, and sulfide can be methylene blue active. Common constituents of sewage and effluents including urea, ammonia, and nitrate as well as the preservatives, formaldehyde and mercury (II) chloride have been shown to give no interference. Nevertheless, not all natural interferents can be eliminated, hence the entities detected are more correctly referred to as methylene blue index (MBAS).

10 TEST REPORT

The test report shall include the following information:

a) Reference to the method used in this standard,
b) An identification of the sample,
c) The results and the method of expression used,
d) Any unusual features noted during the determination, and
e) Details of any operations not included in this standard, or regarded as optional.

ANNEX A

(*Foreword*)

COMMITTEE COMOSITION

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Member Secretary
Ku. Kreeti Das
Scientist B (CHD), BIS

Bureau of Indian Standards

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Review of Indian Standards

Amendments are issued to standards as the need arises on the basis of comments. Standards are also reviewed periodically; a standard along with amendments is reaffirmed when such review indicates that no changes are needed; if the review indicates that changes are needed, it is taken up for revision. Users of Indian Standards should ascertain that they are in possession of the latest amendments or edition by referring to the latest issue of 'BIS Catalogue' and 'Standards: Monthly Additions'.

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Amendments Issued Since Publication

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